# IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

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| **Confirmation No.** | : | 1118 |
| **Appln. No.** | : | 10/620,787 |
| **Applicant** | : | John Simard |
| **Filed** | : | 07/15/2003 |
| **TC/A.U.** | : | 1648 |
| **Examiner** | : | Sharon L. Hurt |
| **Docket No.** | : | 1951300-00006 |
| **Customer No.** | : | 45200 |
| **Title** | : | IMMUNOGENIC COMPOSITIONS DERIVED FROM POXVIRUSES AND METHODS OF USING SAME |

## DECLARATION OF ADRIAN ION BOT, PH.D. UNDER 37 CFR §1.132

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

I, Adrian Ion Bot, declare as follows:

1. I am currently Sr. Director of Scientific Management and Acting Director of Translational Medicine at MannKind Corporation. A copy of my curriculum vitae is attached.

2. I have read and am familiar with the Office Action mailed October 29, 2007 pertaining to this application.

3. I understand that in the Office Action mailed October 29, 2007, the Examiner rejected the claims under 35 USC §103(a) as being obvious over a combination of U.S. Patent Publication No. 2002/0009447, Thomson et al. (J. Immunol. 160:1717-1723, 1998).

4. I have been asked to give my scientific opinion as to certain questions as an expert in immunology and as a person familiar with the level of skill in the art. It is my understanding that these questions are related to factual issues that have arisen in the above-referenced patent application. In this Declaration, I will state each question in a numbered paragraph and will provide my scientific opinion relating to that question in one or more numbered and lettered subparagraphs.

5. Does Thomson (*J. Immunol.* 160:1717-1723, 1998) teach or suggest a polyprotein?

5a. No, Thomson neither teaches nor suggests a polyprotein. Thomson never uses the term polyprotein. Thomson does teach a polyepitope or "polytope". The Office Action takes the position that these terms are equivalent or at least overlapping in meaning. This assertion is incorrect and unsupportable. As explained below the two terms are commonly understood by one of skill in the art to have distinct meanings and under any reasonable interpretation refer to macromolecules that are structurally and functionally distinct.

6. What is a polyepitope?

6a. At a basic level a polyepitope is a polypeptide made up of many epitopes. This raises the question: What is an epitope?

6b. The Online Medical Dictionary (http://cancerweb.ncl.ac.uk/omd/) defines epitope as "That part of an antigenic molecule to which the T-cell receptor responds, a site on a large molecule against which an antibody will be produced and to which it will bind". While other sources will undoubtedly vary in the exact phraseology this definition nicely sets forth key elements of how this term is understood by one of skill in the art. Notably it points out that an epitope is only a part of an antigen, specifically that part that interacts with the immune-cell receptor, and that there are distinctions between epitopes

that interact with T-cell receptors and those that interact with B-cell receptors (antibodies).

6c. Thomson, in the abstract, describes his polyepitope as containing "multiple contiguous minimal murine CTL [Cytotoxic T Lymphocyte] epitopes". The article also describes that the multiple epitopes derive from diverse antigens. This is entirely consistent with the general understanding of the term polyepitope to one of skill in the art. This definition along with other details provided in the paper, conveys that the various epitopes are directly joined one to the next (contiguous), that these are T-cell receptor interacting epitopes presented by class I MHC (CTL epitopes), and that the epitopes are limited in size to only the length, generally 8-10 amino acids, that can be effectively bound by class I MHC molecules (minimal).

6d. Thus a polyepitope is an artificial polypeptide in which multiple minimal (T cell) epitopes are arranged adjacent to each other in a continuous string.

7. What is a polyprotein?

7a. As above, at a basic level a polyprotein is a macromolecule made up of at least 2 proteins, and this raises the question: What is a protein?

7b. There are many aspects to what "a protein" means. One of these aspects is a minimum size. Several sources indicate that a protein typically contains at least 100 amino acids (e.g., *Molecular Cell Biology* 5th ed., Lodish et al. W.H. Freeman & Co. New York, p. 9) or is at least 10,000 MW, roughly 90 amino acids, (e.g., *Lehninger Principles of Biochemistry* 3rd ed., Nelson & Cox, Worth Publishers, New York, p.6; *Biochemistry* 2nd ed., Stryer, W.H. Freeman & Co. New York, p. 19). Others include somewhat smaller molecules. By one source (*Molecular Biotechnology* 2nd ed., Glick & Pasternak, ASM Press, Washington, D.C., p.23) a protein is more than 40 amino acids in length. Another (Biochemistry 2nd ed., Lehninger, Worth Publishers, New York, p.58) teaches proteins range upward from 5000 MW, or roughly 45 amino acids. Finally in *Molecular*

*Biology of the Cell* (2nd ed., Alberts et al., Garland Publishing, New York, p. 112) one learns that proteins are made up of one or more structured domains of 50-350 amino acids. Thus although there is not a universally accepted absolute minimum size for a protein it is equally clear that there is an accepted meaning in the field including a size limitation and that a peptide sequence substantially less than 40 amino acids does not conform to the meaning that "a protein" has to one of skill in the art.

7c. Reading the same sources used for the size discussion in 7b above, several other aspects of what "a protein" means also become apparent. A protein is the translation product of a gene and possesses a particular structure. Thus the term also implies a degree of completeness.

7d. Referring to the instant application, we are taught that the polyproteins of the instant invention are made up of two or more proteins or portions thereof ([0008] sentence bridging pages 2-3), those portions particularly retaining the surface-exposed, immunogenic part of the protein (Example 3, Figure 9, [0027], [0037], and the application as a whole).

7e. Thus one of skill in the art would understand that the polyproteins of the instant invention comprise multiple units each consisting of at least about 40 amino acids and comprising, if not the whole amino acid sequence of the antigenic protein to be incorporated into the polyprotein, then at least a surface exposed portion of the antigenic protein retaining antigenic structure.

8. Comparison of sections 6 and 7 above make abundantly clear that the epitopes of Thomson's polyepitope and the proteins of the instant invention's polyprotein are not the same, nor do they overlap. In other words, fragments of an antigen 8-10 amino acids in length are distinctly different from complete or substantially complete antigens of at least 40 amino acids in length. Thus Thomson does not teach the polyprotein of the instant invention.

9. Would a skilled person be motivated to use the disclosure of Thomson to achieve the polyprotein of the instant invention or perceive a suggestion to do so?

9a. Thomson explicitly teaches the use of minimal epitopes so as to maximize the number of epitopes that can be delivered by a single, relatively small DNA construct and denigrates as problematic the use of a single plasmid to express multiple proteins (see the abstract, 1st paragraph of the article, and last paragraph of the Discussion section). Thus Thomson suggests against encoding whole or substantially whole proteins in a single vector as in the polyprotein of the instant invention.

9b. A reading of the instant application indicates that the immunogenic compositions (and component polyproteins) should be capable of inducing an immune response including a neutralizing antibody response (see, for example, [0019], [0025], [0029], [0033], [0036-7], Examples 1, 3, 6, and 7). Antibodies, particularly neutralizing antibodies, are likely to recognize conformational (discontinuous) epitopes which depend on the native 3-dimensional structure of the antigen (see for example, *Fundamental Immunology*, 4th ed. Paul, Lippincott-Raven Publishers, Philadelphia, pp. 657 2nd col. to 661). In contrast CTL epitopes must be processed into short, necessarily continuous peptides and presented by MHC proteins in order to be recognized by the immune system (see for example, *Fundamental Immunology*, 4th ed. Paul, Lippincott-Raven Publishers, Philadelphia, pp. 669-671). For these reasons one of skill in the art would not consider an article such as Thomson, which is directed to vectors for the generation of a CTL response, to teach anything particularly pertinent to designing vectors for the generation of an immune response including a neutralizing antibody response.

9c. Thomson explicitly reports that despite the inclusion of a known, highly immunogenic linear antibody epitope in his construct no antibody response was obtained (paragraph bridging pp. 1719-21; last paragraph, left col. p. 1722). Thus one of skill in the art would consider that Thomson's design was unpromising for the generation of an immune response including a neutralizing antibody response.

9d. Thomson discloses immunization with the plasmid and the polyepitope product is expressed cytoplasmically where it can readily enter the antigen processing pathway leading to class I MHC-restricted presentation s In primary embodiments of the instant invention, the immunogen is the polyprotein itself. The administration of Thomson's polyepitope product directly would not have ready access to the appropriate processing pathway as is the goal in Thomson. Furthermore, dosage volume and viscosity issues promote minimizing the size of plasmids used as vaccines, as alluded to by Thomson. A plasmid encoding a polyprotein having the multivalency of the Thomson vaccine would be difficult, if not impossible, to administer in an effective dosage. These facts further illustrate the lack of interchangeability of the vaccine approaches taught by Thomson and the instant invention.

10. For each and all of these reasons Thomson cannot be modified to suggest the polyprotein of the instant invention.

11. All statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further, that the statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under section 1001 of Title 18 of the United States Code, and such willful false statements may jeopardize the validity of the application or any patents issuing thereon.

Adrian Ion Bot

Date April 29th, 2008

Adrian Ion Bot, Ph.D.

# Adrian Bot, M.D., Ph.D.

## EDUCATION

- **M.D.,** University of Medicine and Pharmacy Timisoara, Romania.
- **Ph.D.,** Biomedical Sciences / Microbiology - Immunology**,** Mount Sinai School of Medicine, New York.

## PROFESSIONAL COURSES

- **Biotechnology Business Development**, Commercializing a discovery, U.C. Berkeley, 2006.
- **Project Team Workshop,** I.C.S. group, 2006.

## PROFESSIONAL EXPERIENCE

**MannKind Corporation,** Valencia, CA (2003-present)

- 2006-present, Senior Director, Translational Medicine
- 2005-2006, Senior Director, Research and Development
- 2003 to 2005, Director of Immunology and Cancer Research

Responsibilities (in brief): oncology R&D

- Lead an R&D division responsible for designing and preclinical development of lead candidates and biomarkers for multiple oncological indications, resulting in INDs.
    - MKC 1106 PP immunotherapy in multiple solid tumors (clinical trials)
    - MKC 1106 MT in melanoma (preclinical development)
- Translational research support of small molecule projects in cancer and inflammation and biomolecules in cancer
- Management of multiple cost centers and diverse projects in U.S. and internationally (+30 FTEs and annual $ +5.5 mill in operating expenses)
- Provided highest level expertise in immunology; interacted with SAB, Executive team and BOD.
- Advised to Alfred Mann Foundation
- Member or leader of multiple therapeutic oriented project teams.
- Member of program strategic and development teams in (inhaled insulin – phase 3 development; pre-NDA and oncology – preclinical, IND and clinical exploration)
- Design / implement and new technology assessment process, resulting in licensing of therapeutic candidates and technologies.

**AlleCure Corp.,** Valencia, CA (2002-2003)

- 2002 to 2003, Director of Immunology Research at AlleCure Corp., Valencia – CA; acquired by MannKind Corporation

Responsibilities (in brief):

- Leading a team of scientists and associates in evaluating lead candidates for inflammatory and infectious diseases.
- Manage a yearly research budget of + $1 mill and up to ten FTEs
- Responsible for new technology evaluation and management support through successful merger acquisition

**Alliance Pharmaceutical Corp**., San Diego, CA (1998-2002)

- 2001 to 2002, Director of Immunology Research
- 1999 to 2001, Principal Scientist, Department of Exploratory Biological Research
- 1998 to1999; Research Fellow, Department of New Technology Assessment

Responsibilities (in brief):

- Preclinical evaluation (rodents and primates) of a DNA vaccine against influenza virus
- Evaluation, in-licensing of new biotherapies such as recombinant Ig and dsRNA motifs for inflammatory diseases, influenza virus and cancer;
- Evaluation of novel particles for inhaled delivery of biotherapeutics and support on development of spray dried microparticles licensed by Nektar – currently in phase III trials with Tobramycine in Cystic Fibrosis (Chiron)
- Research new adjuvants (synthetic TLR ligands) licensed by MultiCell and currently evaluated preclinically for avian flu; recombinant immunoglobulins licensed and evaluated for type 1 diabetes, MS and celiac disease

**Scripps Research Institute**, La Jolla, CA (1998-1999)

- Guest Scientist, Department of Neuropharmacology, Division of Virology

Responsibilities (in brief): evaluation of transcription factors in anti-viral immune response and inflammatory diseases

**Mount Sinai School of Medicine**, New York, NY (1994-1998)

- Graduate Student, Department of Microbiology

Responsibilities (in brief):

- Preclinical evaluation of a new generation of vaccines (DNA vector based); mechanisms of immunity against influenza virus

- DNA vaccines licensed by Alliance Pharmaceutical Corp.

**University of Medicine and Pharmacy**, Timisoara, Romania (1993-1994)

- Assistant Professor, Department of Physiology.

## EDITORIAL AND REVIEWER ACTIVITIES

Editor in Chief

*International Reviews of Immunology* 2005-present

Associate Editor

*Journal of Immunology (2001-2005)*

Reviewer:

*Transplantation*
*Journal of Immunology*
*Expert Opinion on Drug Delivery*
*Autoimmunity*
*Journal of Leukocyte Biology*
*Vaccine*
*Clinical and Experimental Immunology*
*Viral Immunology*
*Cellular Immunology*

## APPOINTMENTS AND ACTIVITIES

Scientific Advisory Board, Praxair (Fortune 300 company) 2007- present

Scientific Advisory Board of the Cancer Vaccine Consortium (Sabin Vaccine Institute) 2005 - present

Scientific Advisory Board of Astral Inc., San Diego CA (2002-2004)

Scientific Advisory Board, MultiCell Therapeutics, San Diego CA (2005-2007)

Chair, R&D Symposium of MannKind Corporation (Westwood, CA) 2005

Member, Cancer Vaccine Consortium Clinical Trials Working Group 2006

Member, Gerson Lehrman Group Councils 2006-present

Proficiency Panel Program ICS, ELISPOT and Tetramer / Pentamer Staining (Cancer Vaccine Consortium, Sabin Vaccine Institute) 2007

Panel: Southern California Biomedical Council – 'Are we closer to developing a true cancer vaccine?', University of Southern California, Davidson Center, August 30th, 2007

Moderator, Translational Medicine Session, Next Generation Pharmaceuticals Summit, Scottsdale – AZ, Sep 5-7th, 2007

## PROFESSIONAL MEMBERSHIP

Cancer Vaccine Consortium

The American Association for the Advancement of Science

The American Association of Immunologists (past)

The Alumni Association of the Mount Sinai School of Medicine and City University of New York

## PUBLICATIONS - RESEARCH ARTICLES:

1. Weber, J., Boswell, W., Smith, J., Hersh, E., Snively, J., Diaz, M., Miles, S., Liu, X., Obrocea, M., Qiu, Z., and Bot, A. Phase I trial of intranodal injection of a Melan-A / MART-1 DNA plasmid vaccine in patients with stage IV melanoma. **Journal of Immunotherapy**, in press.

2. Smith K, Tam V, Qiu Z and Bot A. Multivalent cellular immune responses detected by iTAG™ tetramers. **Cellular Research** (Beckman Coulter), 1:4-5 (2007).

3. Bot A, Smith D, Phillips B, Bot S, Bona C, Zaghouani H. Immunologic control of tumors by in vivo Fc gamma receptor-targeted antigen loading in conjunction with double-stranded RNA-mediated immune modulation. **J Immunol.** 176(3):1363-74 (2006).

4. Johansen P, Senti G, Martinez Gomez JM, Storni T, von Beust BR, Wuthrich B, Bot A, Kundig TM. Toll-like receptor ligands as adjuvants in allergen-specific immunotherapy. **Clin Exp Allergy.** 35(12):1591-8 (2005).

5. Johansen P, Senti G, Martinez Gomez JM, Wuthrich B, Bot A, Kundig TM. Heat denaturation, a simple method to improve the immunotherapeutic potential of allergens. **Eur J Immunol.** 35(12):3591-8 (2005).

6. von Beust BR, Johansen P, Smith KA, Bot A, Storni T, Kundig TM Improving the therapeutic index of CpG oligodeoxynucleotides by intralymphatic administration. **Eur J Immunol**., 35:1869-76 (2005).

7. P. Johansen, A.C. Haffner, F. Koch, K. Zepter, I. Erdmann, K. Maloy, J.J. Simard, T. Storni, G. Senti, A. Bot, B. Wuthrich and T.M. Kundig. Direct intralymphatic injection of peptide vaccines enhances immunogenicity. **Eur J Immunol.,** 35:568-574 (2005).

8. L. Dellamary, D.J. Smith, A. Bloom, S. Bot, G.R. Guo, H. Deshmuck, M. Costello and A. Bot. Rational design of solid aerosols for immunoglobulin delivery by modulation of aerodynamic and release characteristics. **Journal of Controlled Release**, 95: 489-500 (2004).

9. D.J. Smith, S. Bot, L. Dellamary and A. Bot. Evaluation of novel aerosol formulations designed for mucosal vaccination against influenza virus. **Vaccine**, 21:2805-2812 (2003).

10. A. Bot, E. Rodrigo, T. Wolfe, S. Bot and M.G. von Herrath. Infection-triggered regulatory mechanisms override the role of STAT 4 in control of the immune response to influenza virus antigens. **Journal of Virology**, 77(10): 5794-5800 (2003).

11. L. Wang, Smith D., Bot S., Dellamary L., Bloom A., and Bot A. Non-coding RNA danger motifs bridge the innate with adaptive immunity and are potent adjuvants for mucosal vaccination. **Journal of Clinical Investigation**, 110:1175-1184 (2002).

12. L. Wang, Bot. S., Smith D., Guo G.R., Phillips B., Dellamary L., and Bot, A. Basic immunological properties of spray-dried microparticles developed for nasal and systemic administration of vaccines. **S.T.P. Pharma Sciences** 12:53-61 (2002).

13. T. Wolfe, Bot A., Hughes A, Mohrle U, Rodrigo E, Jaume JC, Baekkeskov S, and von Herrath M. Endogenous expression levels of autoantigens influence success or failure of DNA immunizations to prevent type 1 diabetes: addition of IL-4 increases safety. **European Journal of Immunololy** 32:113-121 (2002)

14. A. Bot, Smith D, Bot S, Hughes A, Wolfe T, Wang L, Woods C, and von Herrath M. Plasmid vaccination with insulin B chain prevents autoimmune diabetes in nonobese diabetic mice. **Journal of Immunology** 167:2950-2955 (2001)

15. A. Bot, Smith DJ, Bot S, Dellamary L, Tarara TE, Harders S, Phillips W, Weers JG, and Woods CM. Receptor-mediated targeting of spray-dried lipid particles coformulated with immunoglobulin and loaded with a prototype vaccine. **Pharmaceutical Research.** 8:971-979 (2001).

16. D.L. Radu, Antohi, S., Bot, A., Miller, A., Mirarchi, A., and Bona, C. Effect of maternal antibodies on influenza virus-specific immune response elicited by

inactivated virus and naked DNA. **Scandinavian Journal of Immunology** 53:475-82 (2001).

17. A. Bot, Shearer, M., Bot, S., Avriette, M., Garcia-Sastre, A., White, G., Woods, C., Kennedy, R., and C.A. Bona. Induction of immunological memory in baboons primed with DNA vaccine as neonates. **Vaccine**. 19:1960-1967 (2001).

18. A. Bot, Stan, A.-S., Inaba, K., Steinman, R., and C.A. Bona: Dendritic cells at a DNA vaccination site express an encoded influenza nucleoprotein and prime CD8+ cytolytic lymphocytes upon adoptive transfer. **International Immunology**, 12:825-832, (2000).

19. A. Bot, Holz, A., Wolfe, T., Temann, A., Flavell, R., and M.G. von Herrath. Local IL-4 expression in the lung reduces pulmonary influenza virus specific secondary cytotoxic T cell responses. **Virology**, 269:66-77 (2000).

20. A. Bot, Tarara, T., Smith, D., Bot, S., Woods, C., and J. Weers. Novel lipid-based hollow-porous microparticles as a platform for immunoglobulin delivery to the respiratory tract. **Pharmaceutical Research**, 17:274-283 (2000) .

21. A. Bot, Shearer, M., Bot, S., Garcia-Sastre, A., Woods, C., Limmer, J., Kennedy, R., Casares, S., and C. Bona. Induction of antibody response by DNA immunization of newborn baboons against influenza virus. **Viral Immunology**, 12:91-96 (1999).

22. Homan, D., Holz, A., Bot, A., Coon, B., Wolfe, T., Petersen, J., Dyrberg, T.P., Grusby, M.J., and von Herrath, M.G. Autoreactive CD4+ lymphocytes protect from autoimmune diabetes via bystander suppression using the IL-4/STAT-6 pathway. **Immunity**, 11:463-472 (1999).

23. Holz, A., Bot, A., Wolfe, T., Grusby, M.J., and M.G von Herrath. Essential role for the IL-12/STAT-4 signaling pathway in autoimmune diabetes. **Journal of Immunology**, 163:5374-5382 (1999).

24. Radu, D.L., Antohi, S., Bot, A., Weksler, M.E., and Bona, C. Plasmid expressing the influenza HA gene protects old mice from lethal challenge with influenza virus. **Viral Immunology**, 12:217-226 (1999).

25. A. Bot, S. Bot and C.A. Bona: Enhanced protection following neonatal immunization with a combination of plasmids expressing NP and HA of influenza virus. **Vaccine**, 16:1675-1682 (1998).

26. A. Bot, Bot, S., and C.A. Bona: Protective role of interferon-gamma during recall responses to influenza virus. **Journal of Virology**, 72:6637-6645 (1998).

27. A. Bot, Bot, S., Garcia-Sastre, A., and C.A. Bona: Protective cellular immunity against Influenza virus induced by plasmid inoculation of newborn mice. **Developmental Immunology**, 5:197-210 (1998).

28. A. Bot, S. Casares, S. Bot, H. von Boehmer and C.A. Bona: Cellular mechanisms involved in protection from influenza virus infection in transgenic mice expressing a TCR receptor specific for class II hemagglutinin peptide in CD4+ and CD8+ T cells. **Journal of Immunology**, 160:4500-4507 (1998).

29. S. Antohi, Bot, A., Manfield, L., and C.A. Bona: Hemagglutinin specific clonotype reactivity pattern of mice immunized as neonates or adults with naked DNA. **International Immunology**, 10:651-662 (1998).

30. A. Bot, Antohi, S., Bot, S., Garcia-Sastre, A., and C.A. Bona: Induction of humoral and cellular immunity against influenza virus by immunization of newborn mice with a plasmid bearing a hemagglutinin gene. **International Immunology**, 9:1641-1650 (1997).

31. S. Casares, Brumeanu, T.-D., Bot, A., and C.A. Bona: Protective immunity elicited by vaccination with DNA encoding for a B cell and a T cell epitope of the A/PR/8/34 influenza virus. **Viral Immunology**, 10:129-136 (1997).

32. T.D. Brumeanu, S. Casares, A. Bot, S. Bot, and C.A. Bona: Immunogenicity of a contiguous T-B synthetic epitope of the A/PR8/34 influenza virus. **Journal of Virology**, 71:5473-5480 (1997).

33. T.D. Brumeanu, Bot, A., Bona, C.A., Dehazya, P., Wolf, I., and H. Zaghouani: Engineering of doubly antigenized immunoglobulins expressing T and B viral epitopes. **Immunotechnology**, 2:85-95 (1996).

34. A. Bot, Reichlin, A., Isobe, H., Schulman, J.,Yokoyama, W. M., and C.A.Bona: Cellular mechanism involved in protection and recovery from influenza virus infection in immunodeficient mice. **Journal of Virology**, 70:5668-5672 (1996).

35. A. Bot, Nangpal, A., Pricop, L., Bogen, B., Kaushik, A., and C.A. Bona: V□-light chain genes reconstitute immune responses to defined carbohydrate antigens or haptens by utilizing different VH genes. **Molecular Immunology**, 33: 1359-1369 (1996).

36. A. Bot, S., Garcia-Sastre, A., and C.A. Bona: DNA immunization of newborn mice with a plasmid expressing nucleoprotein of influenza virus. **Viral Immunology**, 9:207-210 (1996).

37. A.Bot, Bot, S.,Antohi, S., Karjalainen,K., and C.A. Bona: Kinetics of generation and persistence on membrane class II molecules of a viral peptide expressed on foreign and self proteins. **Journal of Immunology**, 157:3436-3442 (1996).

**PUBLICATIONS - REVIEWS**

1. A. Bot, Antohi, S., and C.A. Bona: Immune response of neonates elicited by somatic transgene vaccination with naked DNA. **Frontiers in Bioscience**, 2:173-188 (1997).

2. A.Bot: Immunoglobulin deficient mice generated by gene targeting as models for studying the immune response. **International Reviews of Immunology**, 13:327-340 (1996).

3. C.A. Bona and A. Bot: Neonatal immunoresponsiveness. **The Immunologist**, 5/1:5-9 (1997).

4. A. Bot, Isobe, H., and C.A. Bona: Immunodeficient mouse models in the characterization of the protective immunity to influenza virus. **Folia Microbiologica**, 43:477-478 (1998).

5. S. Casares, Bot, A., Brumeanu, T.-D., and C.A. Bona: Foreign peptides expressed in engineered chimeric self molecules. **Biotechnology and Genetic Engineering Reviews**, 15:159-198 (1998).

6. A. Bot: DNA vaccination and the immune responsiveness of neonates. **International Reviews of Immunology**, 19:221-245 (2000).

7. A. Bot, and Bona C. Genetic immunization of neonates. **Microbes and Infection** 4:511-20 (2002).

8. A. Bot, W. Phillips and M.G. von Herrath. Antigen-based immune modulation: DNA vectors and beyond. **Expert Opinion in Biological Therapeutics**, 2(8):929-42 (2002).

9. M.G. von Herrath, and A. Bot. Immune responsiveness, tolerance and dsRNA: implications for traditional paradigms. **Trends in Immunology**, 24(6): 290-293 (2003).

10. A. Bot, K. Smith, and M.G. von Herrath. Molecular and cellular control of T1/T2 immunity at the interface between antimicrobial defense and immune pathology. **DNA and Cell Biology**, 23(6):341-350 (2004).

11. A. Bot and M.G. von Herrath. Prophylaxis of autoimmune diabetes by antigen based immune modulation: are we there yet? **Minerva Med**. 95(2):105-14 (2004).

12. A. Bot. EDITORIAL. **International Reviews of Immunology**. 24(5):285-6 (2005).

13. W.J. Phillips, D.J. Smith, C.A. Bona, A. Bot and H. Zaghouani. Recombinant immunoglobulin-based epitope delivery: a novel class of autoimmune regulators. **International Reviews of Immunology**. 24(5):501-17 (2005).

14. A. Bot. The spectrum of influenza virus pandemic: between agony and hope. **Timisoara Medical Journal**, 56(1), 7-14 (2006).

## BOOKS AND CHAPTERS

1. **Genetic Immunization**; by C.A. Bona and A. Bot. *Kluwer Academic / Plenum Publishers* - New York, Boston, Dordrecht, London, Moscow - 2000 (179 pages; ISBN 0-306-46226-5).

2. A. Bot and D. Smith. Fc receptor targeting with recombinant immunoglobulins and immunoglobulin formulations. In **Cellular Drug Delivery – Principles and Practice**, *Eds. R. Lu and S. Øie*, **Humana Press**, 287-310 (2004).

3. M.G. von Herrath and A. Bot. Plasmid-mediated delivery of antigens or biological response modifiers as means to suppress autoimmunity. In **Treatment of Autoimmune Disorders**, *Eds. M. Sticherling and E. Cristophers*, **Springer-Verlag**, Wien - New York, 151-165 (2003).

4. C.A. Bona, Bot A., and T.-D. Brumeanu: Immunogenicity of viral epitopes expressed on genetically enginereed immunoglobulins. In *Antibody Engineering*, ed. J.D. Capra, Basel, Karger; **Chemical Immunology**, 65:179-206, (1997).

5. A. Bot, and C.A. Bona. Mechanisms of protection against influenza virus. In **Recent Research Developments in Virology** - **Transworld Research Network**, 1:317-325, (1999).

6. Eds. A. Bot and M. Obrocea. **Challenges and Opportunities in the Translation of Cancer Vaccines, Informa Healthcare**, in press.

7. A. Bot and M. Obrocea, Bidirectional Bedside Lab Bench Processes and Flexible Trial Design as a Means to Expedite the Development of Novel Immunotherapeutics, in **Challenges and Opportunities in the Translation of Cancer Vaccines, Informa Healthcare**, *Eds. A. Bot and M. Obrocea*, in press.

## INVITED LECTURES

1. **International Society for Biological Therapy of Cancer** November 10-13, 2005, Alexandria, VA ; "A novel class of biotherapeutics co-targeting cancer cells and the associated tumor neovasculature".

2. **Mount Sinai School of Medicine** - Microbiology Seminars, May 9th 2006, "Improving on the therapeutic index of synthetic dsRNA: a new class of potent adjuvants"

3. **6th Annual Meeting of the Cancer Vaccine Consortium**, November 10-11, 2006, Washington DC; "Translational Approach in Support of Clinical Development".

4. **University of Southern California,** Norris Cancer Center, February 9th 2007, Los Angeles. "Innovative Approaches to Target Major Histocompatibility Molecule - Peptide Complexes onto Tumor Cells"

## ORAL ABSTRACTS

1. A. Bot, K. Smith, Xiping Liu, Liping Liu, Z. Qiu, T. Kuendig: Potent immunity achieved by targeted, sequential administration of recombinant DNA vectors and anchor-modified epitope peptides, **International Society for Biological Therapy of Cancer** November 10-13, 2005, Alexandria, VA (J. Immunother vol. 28, no. 6, 2005)

2. A Bot, K.A. Smith, X. Liu, and Z. Qiu. *In situ* targeting of antigen presenting cells within secondary lymphoid organs as a means to control immune responses. **International Society for Biological Therapy of Cancer** November 10-13, 2005, Alexandria, VA (J. Immunother vol. 28, no. 6, 2005)

3. Kent A. Smith, Zhiyong Qiu, Ani Der-Sarkissian, Robb Pagarigan, Brenna Meisenburg, Victor Tam, Gene Girgis, Thomas M. Kündig, W. Martin Kast, and Adrian Bot. Immunological Control and Regression of Solid Tumors Achieved by Lymph Node Targeted Immunotherapy. **International Society for Biological Therapy of Cancer**, *November 26-29, 2006, Los Angeles, CA* (J. of Immunother., Vol 29, No 6, 676-677, 2006).

4. Zhiyong Qiu, Kent A. Smith, Ani Der-Sarkissian, Victor Tam, and Adrian Bot. Plasmid-driven Antigen Expression is Essential during Priming of Immune Response using a Potent Intra Lymph Node Immunization Approach. **DNA Vaccines** 2007, Malaga, Spain, May 23-26, 2007.

## POSTERS AND ABSTRACTS

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## AWARDS, HONORS AND GRANTS

*Patents awarded*

1. A. Bot and C. Bona. Immunization of infants. **United States Patent 6,204,250** Issued on March 20, 2001; Filed on November 22, 1996.

2. A. Bot, T. Tarara, J. Weers, A. Kabalnov, E.G. Schutt, and L.A. Dellamary. Particulate delivery systems and methods of use. **United States Patent 6,630,169**. Issued on October 7, 2003. Filed on December 22, 2000.

3. Bot, Adrian I.; Dellamary, Luis A.; Smith, Dan J. Methods and compositions for delivering macromolecules to or via the respiratory tract **United States Patent 7,141,236**. Issued on 2006-11-28. Filed on 2002-04-26.

*Grants*

1998. **Research Grant:** "Genetic Immunization of Neonates against Influenza Virus". Co-Principal investigator: Adrian Bot, MD, PhD. Agency: National Institutes of Health, United States of America. Awarded (active September 1998- August 1999).

1999. **Research Grant:** "Down-regulation of diabetogenic T cells by chimeric antigen presenting molecules" Co-investigator: Adrian Bot, MD, PhD. Agency: National Institutes of Health, United States of America. Awarded (active April 2000 - March 2001).

2000. **Research Grant:** "Treatment of EAE using novel delivery systems". Co-Principal Investigator: Adrian Bot, MD, PhD. Agency: National Institutes of Health, United States of America. Awarded (active September 2000- August 2001).

2000. **Research Grant:** "Formulation for mucosal immunization against HIV". Principal Investigator: Adrian Bot, MD, PhD. Agency: National Institutes of Health, United States of America. Awarded (active July 2000 - June 2002).

*Miscellaneous*

1986. Silver Medals (Team and Individual) at the **XVII International Olympiad of Physics,** London - Harrow on the Hill, United Kingdom.

1989. The National Student Award for Medical Physiology: Second Place; Bucharest, Romania.

1993. Graduate student fellowship granted by the Mount Sinai School of Medicine of the City University of New York, for the period of 1994-1998.

## PATENT APPLICATIONS (US Patent and Trademark Office)

1 A. Bot, L. Dellamary, D. Smith, and C.M. Woods. Engineered spray-dried lipid-based microparticles for cellular targeting. **United States Patent Application 20020103165**.

2 A. Bot and C. Bona. Immunization of infants. **United States Patent Applications 20020115625, 20020103145, and 20040053871.**

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